

**Project Title:**

Methyl Bromide Alternatives Research for Michigan  
Herbaceous Perennial Ornamental Production

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Final Report

**Proposal Synopsis**

The Michigan herbaceous perennial ornamentals industry has relied on methyl bromide for nematode and weed management. This product is being phased-out under the Montreal Protocol. There is a distinct need for highly effective alternative nematode and weed management procedures. From the fall of 2002 through the fall of 2004, a 3.2 acre USDA-funded methyl-bromide alternatives trial was conducted at Sawyer Nursery in Hudsonville. Several potential alternatives were identified and the industry requested that their efficacy and economics be demonstrated again at a second location under the conditions of different growing seasons. Methyl Bromide Alternatives Research for Michigan Herbaceous Perennial Ornamental Production was designed to provide 2005-06 resources for the requested research. Ponderosa Nursery in Hamilton, Michigan was used as the research site. Although this is funded as a one-year project (August 1, 2005 through July 31, 2006) the work started in 2004 and will continue the 2006 and 2007 growing seasons as an integral component of the USDA-funded projects.

**Impact on Michigan Nursery/Ornamental Horticulture Industry:**

Nematodes and weeds are key pests in herbaceous perennial ornamental production in Michigan. With the phase-out of methyl bromide under the Montreal Protocol, it is imperative that effective and economically viable alternatives be identified and demonstrated to the producer community. The results of this research and two other associated projects demonstrate that alternatives for methyl bromide under Michigan growing conditions are going to be site or enterprise specific. Weed and nematode population pressures at Ponderosa Nursery were considerably higher than those at Sawyer Nursery, resulting in significantly greater challenges related to weed and nematode control in the absence of methyl bromide. The project was implemented as part of

both the first (2002-2005) and second (2004-2007) Michigan herbaceous and woody perennial ornamental methyl bromide alternatives research grants obtained from USDA/CSREES.

### **Objectives and Hypotheses:**

The objectives of this applied research initiative were to:

- 1) develop and demonstrate alternatives to methyl bromide for the control of plant-parasitic nematodes and weeds for the MI herbaceous perennial ornamentals industry and
- 2) make the results of this project available to the MI herbaceous perennial ornamentals industry through field days, trade publications and a specific mailing of the final reports for both this project and the original USDA research work.

The following hypotheses were tested:

- Telone II applied under tarp at 35 gallons per acre will provide northern root-knot nematode (*Meloidogyne hapla*) and broad-spectrum/long-term weed control that is comparable to that of methyl bromide (98% MeBr, 2% chloropicrin) applied under tarp at 400 pounds per acre in Hosta and Silver Mound production initiated with high quality (northern root-knot nematode-free) propagation stock.
- Applications of Vydate, and Chancellor WD or BioNem will control bulb and stem nematodes (*Ditylenchus dipsaci*) associated with Creeping Phlox grown from general quality (low level of nematode infection) propagation stock grown in either methyl bromide treated or non-fumigated soil under field conditions.
- Applications of Vydate and Chancellor control northern root-knot nematodes associated with Creeping Phlox and Ajuga grown from general quality (low level of nematode infection) propagation stock in either methyl bromide treated or non-fumigated field soil.
- Application of high quality compost will reduce risk to the northern root knot nematode to an acceptable level in Hosta and Silver Mound production initiated with high quality (northern root-knot nematode-free) propagation stock
- The Michigan herbaceous perennial ornamentals industry will adopt alternative nematode and weed management procedures that they have seen demonstration at one or more different commercial production locations.
- It was not possible to test the following hypothesis because of constraints of the experimental design and the lack of registered herbicides. A system of pre-plant and post-plant conventional herbicides was proposed. to provide broad-spectrum weed control that is comparable to that of methyl bromide (98% MeBr, 2% chloropicrin) in Hosta and Silver Mound production initiated with high

quality (northern root-knot nematode-free) propagation stock. This was not possible because of limitations of the experimental design and lack of registered materials.

### Research Methodology

The research consisted of two field trials at Ponderosa Nursery in Hamilton, Michigan. Trial 1. was designed to evaluate root-knot nematode and weed control on Hosta and Silver Mound. Trial 2. was designed to evaluate root, knot nematode, bulb and stem nematode and weed control on Creeping Phlox and Ajuga. Both sites were sampled and staked for the plots for the first time on October 7, 2004. Both sites were monitored extensively in 2005 and 2006 and are not yet ready for plant quality data observations and grower field days. The project, however, was presented at nursery grower meetings in January, February and August 2006. A comprehensive research summary of the first USDA project was distributed at the meeting in February (Appendix A.) and new cover crop and bio-fumigation technology was demonstrated at the meeting in August. As a result, several individuals have requested consultations for developing 2007 plans for implementation of the technology in their enterprises.

Trial 1. consists of five treatments (Table 1), each replicated four times (Figure 1.). The site was fumigated with 350 lbs/A methyl bromide 67/33 for the first time on October 7, 2004. The site was sampled and refumigated with 350 lbs 67/33 methyl bromide for on May 26, 2005. It was also fumigated with 35 gallons Telone II per acre and trapped on this date and tilled on July 27, 2005. Trial I was planted on August 5, 2005, with four replications of clean stock, Hosta (lancifolia) and silver mound (artemisia) one row each and treated with Chancellor Bio-Nema at 4.43 oz per acre in 47 gallons water per acre was also applied on this date. The plot was hand-weeded throughout the 2006 growing season. Missing plants were replanted in the spring and four applications of Chancellor WD were applied throughout the 2006 growing season.

Table 1. Experimental design for methyl bromide alternatives Trial No. 1 at Ponderosa Nursery, Hamilton, Michigan .

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#### Treatments:

- 1 Control (cultivated and hand weeded)
  - 2 Compost (4 tons per acre)
  - 3 Chancellor WD and BioNem
  - 4 Methyl bromide-Chloropicrin (67-33, 3350 lbs. per acre tarped)
  - 5 Telone II (35 gallons per acre tarped)
- 

Figure 1. .Plot Design

301 1	302 4	303 3	304 5	305 2	401 5	402 1	403 2	404 4	405 3
101 5	102 4	103 2	104 3	105 1	201 4	202 2	203 1*	204 5	205 3

Trial 2. consists of 10 treatments (Table 2), each replicated four times (Figure 2). Half of Trial 2. was fumigated with methyl bromide (m) and the other half retained as a control (c). Five treatments each replicated four times were superimposed on this design (Figure 2). The trial was sampled and fumigated and staked on May 26, 2005, and tilled on July 27, 2005. Environmental conditions were far too dry to initiate the research at this time. Compost at 4 tons per acre was applied and incorporated August 4, 2005, starting the irrigation within 15 minutes of the bio-nematicide treatment. Trial 2. was fumigated again with 400 lbs 67/33 methyl bromide per acre September 6, 2005. This was done because it had been worked and reworked to the point that the fumigated plots were very likely contaminated with both nematodes and weed seeds. Trial 2. was planted and treated with Vydate 2L post plant at 2 lbs. a. i. per acre in 25 gallons water per acre. Both Vydate and Chancellor WD were applied throughout the 2006 growing season.

Treatment:

woods

[illegible]

## Results

Weed and nematode pressures at the Ponderosa Site were very high and much more challenging than at Sawyer Nursery. Weed control in Trial 1 was unsatisfactory with both methyl bromide 67/33 and Telone II (Table 3). Nematode control assessment is still in progress and plant growth and quality determinations will not be made until the spring of 2007.

Table 3. Spring 2006, weed indices (0-5, 0 = excellent control, 5 = poor control).

Treatment	Weed Index
Control	4.5 a
Compost	5.0 a
Chancellor Bio-nem	5.0 a
Methyl bromide	4.5 a
Telone II	4.5 a

In Trial 2. weed control with methyl bromide 67/33 was highly variable and not equal to that obtained by the industry on a commercial basis with methyl bromide 98/2 (Table 4). ). Nematode control assessment is still in progress and plant growth and quality determinations will not be made until the spring of 2007.

Table 4. Spring 2006, weed indices (0-5, 0 = excellent weed control, 5 = poor weed control).

Treatments	Weed Index
Non-fumigated Control	4.75 a
Non-fumigated Compost	3.75 a
Non-fumigated Chancellor WD	4.25 a
Non -Fumigated Chancellor	4.75 a
Non-fumigated Vydate	4.75 a
Fumigated Control	1.75 a
Fumigated Compost	2.25 a
Fumigated Chancellor Wc	2.75 a
Fumigated Chancellor	1.75 a
Funigated Vydate	2.00 a

## Summary

No single alternative currently exists for replacement of methyl bromide in the Michigan herbaceous perennial nursery industry. Methyl Iodide worked well when applied at the proper rate at the Sawyer Nursery Trial, but it does not appear that it will be registered. Telone II worked under tarp worked under low weed and nematode pressures, but not under high population densities. Recommendations for weed and nematode management in this industry will have to be site specific for the near future. This is a major change from the use of methyl bromide which provided excellent broad spectrum control of numerous pest species. There is currently, however, a significant amount of interest in the industry to research alternatives such as cover crops and bio-fumigants. This will not be easy, but the technology exists. Before it becomes adopted by the Michigan industry it will have to be refined and adopted on a commercial basis by at least one major enterprise.

**Appendix A.**  
**METHYL BROMIDE I ALTERNATIVES RESEARCH REPORT**  
**WITH SPECIAL REFERENCE TO METHYL BROMIDE**  
**ALTERNATIVES II AND III**

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and B. Zandstra<sup>7</sup>**

**Michigan Methyl Bromide Alternatives Conference**  
**Grand Rapids, Michigan**  
**February 15, 2006**

This research report is designed to summarize the results of the 2003-2005 USDA CSREES Methyl Bromide Alternatives research grant awarded jointly to Michigan State University, Cornell University and the University of Rhode Island. The report begins with an overview of the methyl bromide situation that was prepared for the Michigan Farm Bureau and conclude with information about the 2004-2006 USDA CSREES Methyl Bromide Alternatives research grant awarded to Michigan State University and a methyl bromide alternatives research grant awarded to G. W. Bird by the Michigan Department of Agriculture as part of the Michigan Horticultural Fund. The report for the first methyl bromide research grant is divided into sections on nematode and weed management.

### **Methyl Bromide Overview**

Methyl Bromide (CH<sub>3</sub>Br) is a highly toxic gas. During the past four decades, it has been used widely as a fumigant for control of insect, nematode, fungal and weed pests associated with soil, harvested grain/fruit/vegetables, and structures/equipment. It is often formulated as a mixture with chloropicrin (tear gas, trichloronitromethane). The most important advantages of methyl bromide (MeBr) include its broad spectrum toxicity, high vapor pressure allowing diffusion into relatively non-accessible locations, cost effectiveness and comparatively short pre-plant soil application intervals. A total of 25,528 metric tons of MeBr were used for fumigation purposes in the United States in 1991.

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MeBr is a Class 1. Stratospheric Ozone-Depleting Chemical under the provisions of the U.S. Clean Air Act of 1990<sup>2</sup>. In accordance with this legislation, MeBr production was capped at 1991 levels and its use to be phased-out by 2001. The legislation was amended in 1998 and the phase-out of MeBr placed on the schedule approved for the signatories of the Montreal Protocol. This international treaty mandates that high income nations attain a 25% reduction in MeBr production and importation by 1999, based on their 1991 base-levels, with 50%, 75% and 100% reductions required for 2001, 2003 and 2005, respectively. The Treaty stipulates an average 1995-1998 MeBr base-line for low and middle income nations, with a gradual phase-out for these countries to be completed by January 1, 2015. There are, however, provisions in the Treaty for continued use of MeBr, including the Critical Use Exemption Clause (CUE), implemented by the MeBr Technical Options Committee and Technical and Economic Assessment Panel of the United Nations Environmental Programme (UNEP). In 2003, U.S. requested a 10,000 ton MeBr

CUE for 2005 and 2006, representing 25.5% of the base. Decision on the request was Tabled by the 185 countries present at the UNEP meeting in Nairobi, Kenya. It will be on the agenda for the March 2004 meeting in Montreal, Canada. If approved, licensing requirements, fees and accountability procedures will be part of the CUE process.

A total of 486,972 lb of MeBr was used in Michigan in 2000, representing 0.95% of the U.S. base or 1.91% of the 2001 U.S. Montreal Protocol target. In MI agriculture, MeBr is used primarily as a pre-plant soil fumigant in the production of herbaceous perennial ornamentals, woody seedlings and specific vegetable crops. The cost of MeBr application in MI in 2002 was approximately \$1,750 per acre. It increased to close to \$2,000 in 2003. The Quarantine/Pre-shipment Exemption Clause in the Montreal Protocol does not appear to be the solution to the MI Herbaceous Perennial Ornamental and Woody Seedling Industries problem. Although it applies to a significant amount of the plant material entering interstate commerce, based on current MI statutes, it does not apply to intrastate distribution. The industry does not segregate its field production practices into these categories. MeBr is also used in MI as a post-harvest fumigant for agricultural products and for treatment of pest-infested structures and equipment.

The following is a brief summary of the soil fumigation uses (target pests, industry value and estimated usage), with the target pests prioritized in relation to the current understanding of their importance:

Herbaceous Perennial Ornamental Industry

Target Pests: Nematodes, Weeds, Fungi

Acreage Treated: >90%

Woody Ornamental Seedling Industry

Target Pests: Weeds, Fungi, Nematodes

Acreage Treated: Estimated at about 75%



## Specific Vegetable Crops (tomato, pepper, pumpkins and squash)

Targeted Pests: Fungi, Weeds, Nematodes

Acreage Treated: Estimated at about 25%

The MI MeBr usages are included in the U.S. CUE request for 2005 and 2006. A MI MeBr Task Force was organized in the winter of 2001. This initiative resulted in the successful acquisition of two USDA/CSREES/MeBr Alternatives Program grants. A two-year project was funded in 2002 for a total of \$213,434, dealing primarily with MeBr alternatives for the Herbaceous Perennial Ornamental Industry. A three-year project was funded in 2003 for a total of \$370,701. It focuses on MeBr alternatives for the Woody Seedling Industry. During the four years, the MI MeBr Task Force sponsored a number of educational events related to this project.

A comprehensive review of the status of MeBr alternatives was published in 2003 (Martin, F. N. 2003. *Development of alternative strategies for management of soilborne pathogens currently controlled with methyl bromide*. Ann. Rev. Phytopathol. 41:325-350). Alternatives to MeBr include currently registered fumigants (chloropicrin, 1,3-dichloropropene, methyl isothiocyanate), potential alternative fumigants (methyl iodide, propargyl bromide, ozone), non-fumigant pesticides (herbicides, fungicides, nematicides), non-pesticide approaches to pest management and soil quality enhancement. Although a significant number of the above have potential, none have currently been developed to a high enough level to be satisfactory alternatives for MeBr. USDA CSREES.

### **Methyl Bromide Alternatives Research Project No. 1.**

This project began in the fall of 2002 and was completed in 2005. It was Michigan's first methyl bromide alternatives research project and consisted of the following five parts: 1) a 3.2 acre herbaceous perennial ornamentals research trial for northern root-knot nematode (NRKN) and weed control with 15 treatment each replicated six times at the Sawyer Nursery in Hudsonville, MI, 2) microtile research with nematicides for NRKN control on hosta, tomato and strawberry at Geneva, NY, 3) organic acid research for NRKN control on hosta, tomato and strawberry at the University of Rhode Island, 4) nematode biological control research at MSU and 5) a series of producer education programs and visits to the Sawyer Nursery site.

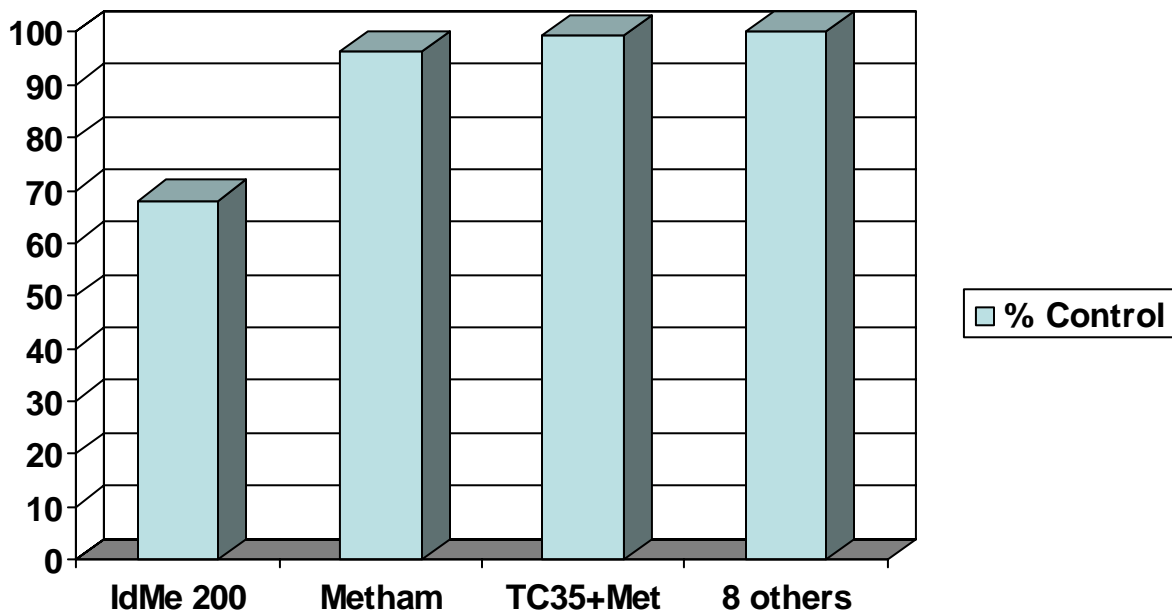
The 3.2 acre multi-year field trial in MI with herbaceous perennial ornamentals was established in the fall of 2002 at Sawyer Nursery in Hudsonville, MI. It consisted of the following 15 treatments:

- |  |           |
|--|-----------|
| 1. Non-treated control (non-tarped)                | 0.00 lb/A |
| 2. Non-treated control (tarped)                    | 0.00 lb/A |
| 3. Idomethane (50%) + 50% chloropicrin (tarped)    | 300 lb/A  |
| 4. Idomethane (50%) + 50% chloropicrin (tarped)    | 200 lb/A  |
| 5. Telone C-35 (tarped)                            | 35 gal/A  |
| 6. Methyl bromide (98%) + 2% chloropicrin (tarped) | 350 lb/A  |

7. Idomethane (98%) + 2% chloropicrin (tarped)	150 lbs/A
8. Metham (not tarped)	75 gal/A (1:4 water)
9. Metham (tarped)	75 gal/A (1:2 water)
10. Metham (tarped)	75 gal/A (1:4 water)
11. Telone II (tarped)	35 gal/A
12. Telone II + Metham (tarped)	35 + 75 gal/A (1:4 water)
13. Methyl bromide (67%) + 33% chlropicrin (tarped)	350 lb/A
14. Telone C-35 + Metham (tarped)	35 + 75 gal/A (1:4 water)
15. Basamid (not tarped)	350 lb/A

The trial was planted to Silver Mound, Blue Glow, Euphorbia, Moon Beam, Snow Lady, Hosta and Munstead on June 23, 2003.

Compared to the non-treated tarped and non-tarped checks, excellent control of plant parasitic nematodes was obtained with all of the treatments except Idomethane (50%) + 50% chloropicrin tarped and applied at 200 lb/A. By the end of the experiment in the fall of 2004, low population densities of plant parasitic nematodes were associated with the three metham and the Telone C-35 + metham treatments.



When the test was terminated in the fall of 2004 and root weights were determined for all six replications of each of the 15 treatments. Crop growth was generally similar among the treatments and controls. Telone C-35 had a negative impact on Euphorbia. Lavender shoot and root growth was negatively impacted by both metham and methyl bromide.

## **New York evaluation of methyl bromide alternative control products against *M. hapla* or *Pratylenchus penetrans* in field microplots:**

The same 96 field microplots (4-ft. diameter, fiberglass cylinders) used for the first year's testing in 2003, were also used for this year's test on tomato, strawberry and hosta. Six treatments, each with 5 replications, were established for each crop: Untreated check, Methyl bromide-fumigated (1 lb/100ft<sup>2</sup>, under plastic), Basamid (granular, 350 lbs./A), Vydate L (2.25 gal. /A), Fosthiazate (4.8 pts. /A), and Agri-Mek (16 ozs/A, 2 applications). An additional treatment of BioYield<sup>TM</sup> {3 g formulation/plant (approx. 2 lbs/cu. yard)} was also included in the tomato test. Tomato plants grown in the methyl bromide-treated plots did not exhibit any root-galling symptoms and eggs of *M. hapla* were not recovered from their roots (Table 1). Plants growing in the methyl bromide-fumigated plots also produced significantly higher number and weight of tomatoes (Table 1). Yield of other treatments did not differ from those produced by the plants growing in the check treatment (untreated, infested plots) (Table 1). However, the root-galling severity ratings and the number of eggs recovered from tomato roots were reduced by the Fosthiazate, Vydate, Basamid, Agri-Mek and BioYield treatments (Table 1). It is not known why a number of the treatments reduced nematode damage and reproduction without affecting yield, although the roots of tomatoes growing in the methyl bromide fumigated plots usually appeared cleaner (white in color) as compared to those in the other treatments. The latter warrant further investigation.

Similarly, total fresh weight of strawberry plants growing in the methyl bromide-treated plots was the highest as compared to that of plants growing in the other treatments (Table 2). Lesion nematodes were not recovered from soils or roots of strawberry plants growing in the methyl bromide- and Basamid-treated plots (Table 2). The number of lesion nematode in roots and soils from the Fosthiazate-treated plots were also very low, and they were also reduced in the Vydate and Agri-Mek treatments as compared to those in the untreated check plots. Marketable yield of strawberries in all the treatment plots will be determined in 2005 growing season.

Results obtained on the same treatments in microplots with hosta last year demonstrated that the variety 'Krossa Regal' was resistant to *Meloidogyne hapla*. However, in a greenhouse test we found that this variety was susceptible to *M. incognita*. In 2004, the hosta variety 'Honeybells' was established in the field microplot test and inoculated with *M. incognita* (race 3 from NC). Unfortunately, symptoms of root-galling were not observed on examined roots and no eggs were recovered from roots. In addition, the hosta plants in all the treatments grew vigorously and there were no differences in top weight or total weight of the plants. No juveniles of the root-knot nematode were recovered from soil of the methyl bromide treated plots when sampled in the fall. Also, the number of juveniles recovered from soils of the other treatments was generally low and showed no differences. Five-hosta cv. 'Honeybells' were inoculated with *M. hapla*, *M. incognita*, or left as non-inoculated checks. Only few root thickenings were observed on the plants inoculated with *M. incognita*. Thus, 'Honeybells' appears to be resistant to *M. hapla* and highly tolerant to *M. incognita*, but additional confirmation of the reaction of both 'Krossa Regal' and 'Honeybells' is warranted.

**Table 1.** Effect of selected control products on growth and yield of tomato and population of *M. hapla*. NYSAES Research Farm microplots, 2004.

<u>Treatment</u>	<u>Total wt/plot</u>		<u>Fruit yield/plot</u>		<u>Root galling</u>	<u>Eggs</u>
	Top(lbs)	Roots(g)	Number	Wt. (lbs)	(1-9)	g/root
untreated	34.2	412	90	21.4	2.1	920
Agri-Mek 0.15 EC	30.8	444	88	18.8	1.4	80
Basamid Granular	34.2	467	105	22.2	1.2	480*
Vydate L	34.7	412	112	23.2	1.4	108
Fosthiazate 900 EC	34.7	416	101	22.7	1.1	0
Methyl Bromide	60.4	614	165	42.0	1.0	0
Bioyield (MeBr fum)	61.1	729	145	39.1	1.0	47
Bioyield	40.5	548	110	26.0	1.5	613
<b>LSD 0.05</b>	<b>11.53</b>	<b>142.5</b>	<b>40.5</b>	<b>9.37</b>	<b>0.39</b>	<b>637.6</b>

**Table 2.** Effect of selected control products on growth of strawberry and population of *P. penetrans*. NYSAES Research Farm microplots, 2004.

<u>Treatment</u>	<u>Weight/3 plants</u>		<u>Root Lesion nematode</u>	
	Total	Roots	per g root	per 100 cc soil
untreated	248	25.2	119	88
Agri-Mek 0.15 EC	207	17.5	86	44
Basamid Granular	256	19.5	0	0
Vydate L	246	21.0	37	44
Fosthiazate 900 EC	256	20.1	3	8
Methyl Bromide	323	23.6	0	0
<b>LSD 0.05</b>	<b>73.9</b>	<b>4.93</b>	<b>68.3</b>	<b>74.0</b>

Although this collaborative project expired at the end of 2004, the established strawberry and hosta microplots (30 of each) were maintained for yield assessment in 2005 without additional treatments. Again, the methyl bromide, Fosthiazate, and basamid soil treatments were highly effective in controlling the lesion nematode (*Pratylenchus penetrans*). However, only the methyl bromide treatment significantly increased the total berry yield of strawberry. Basamid-treated plots resulted in the second highest yield, but it was not significantly different from the check or the other treatments. None of the treatments including the methyl bromide affected the yield (foliage weight) of hosta cv. 'Honeybells', which was growing in *M. incognita* infested plots. Areas of root thickening were observed on only a few roots. In addition, no root-galling symptoms were observed on roots of hosta cv. 'Honeybells' that were uninoculated or inoculated with *M. hapla* or *M. incognita* in a replicated greenhouse test. These results suggest that this cultivar of hosta is highly resistant to both *M. hapla* and *M. incognita*.

### **Rhode Island Development of Organic Acid MeBr Alternatives**

The effect of nematicidal applications of the organic acids butyric acid and propionic acid was investigated against *Meloidogyne hapla* on tomatoes and *Pratylenchus penetrans* on strawberries. Six treatments were employed in 2002 as a baseline. In 2003 and 2004, these rates were reduced to identify the lowest effective concentrations. In both years each treatment was repeated eight times, for an annual total of 96 20,000 cm<sup>3</sup> microplots. *M. hapla* and *P. penetrans* were reared in the greenhouse during the winter of 2001, 2002 and 2003 and inoculated into microplots at a rate of 100,000 *M. hapla* per tomato microplot and 1,500 *P. penetrans* per strawberry microplot in early May. A week later, chemical applications were made to the microplots. Tomatoes and strawberries were transplanted to the appropriate microplots six weeks later, at the end of June. Plants were watered and fertilized regularly throughout the summer and taken down 3 months later, at the end of September. In order to assess nematode pathogenicity and reproduction, the roots and foliage of tomato plants were weighed, root systems were scored for galling severity and roots were subjected to a chlorox extraction to remove eggs, which were then counted. Similarly, roots and foliage of strawberry plants were weighed and nematodes were extracted by placing roots in an erlenmeyer flask on a mechanical shaker for 48 hours.

During the 2002 field season, Concentrations of butyric acid at 1.0M, 0.5M and 0.1M were employed as a preplant nematicidal treatment, in conjunction with an untreated control, a Vydate control and a propionic acid application at 1.0M. Butyric acid rates at 1.0M and 0.5M killed >90% of the plants used, even though they were planted 6 weeks after chemical application. The pH of these plots was approximately 4.5 at planting. Because of the high plant mortality in these microplots, surviving plants were not examined for nematode reproduction, although it is likely that nematode reproduction was minimal. The pH of the Vydate treated plots was 5.81 at planting and the pH of the 0.1M butyric acid treated plots was 5.51 at planting. In the tomato experiment using *M. hapla*, control plants had a significant amount of nematode reproduction with a mean of 73,313 eggs/g root at harvest. Vydate treated plants had a mean of 20,250 eggs/g root and 0.1M butyric acid treated plants had a mean of 17,063 eggs/g root at harvest. The Vydate and butyric acid treatments were statistically similar to each other but statistically different than the control at  $P < 0.001$ . Galling severity showed similar significant differences between treatments (at  $P < 0.001$ ) with a disease index rating of 6.87 for the control plants, 2.81

for the Vydate treated plants and 2.44 for the 0.1M butyric acid treated plants. Only two tomato plants survived the propionic acid treatment but both plants were completely free of nematode galling and reproduction.

In the strawberry experiment using *P. penetrans*, control plants had a significant amount of nematode reproduction with a mean of 286 nematodes/g root. Vydate treated plants had a mean of 66 nematodes/g root and 0.1M butyric acid treated plants had mean of 21 nematodes/g root. The Vydate and butyric treatments were statistically similar to each other but statistically different than the control at  $P<0.001$ .

During the 2003 field season, Concentrations of butyric acid at 0.1M, 0.05M and 0.01M were employed as a preplant nematicidal treatment, in conjunction with an untreated control, a Vydate control and a propionic acid application at 0.05M. In the tomato experiment using *M. hapla*, control plants had a minimal amount of nematode reproduction with a mean of 1,121 eggs/g root at harvest. Nematode reproduction in other treatments ranged from 0 (Vydate) to 109 eggs/g root (0.01M butyric acid). All chemical treatments were statistically similar to each other and statistically different from the control at  $P<0.001$ . Galling severity of control plants was minimal with a disease index rating of 2.5. While all of the chemical treatments applied were highly successful, it is unclear how these treatments would have preformed under high disease pressure. It is also unclear why root-knot nematode populations remained so low, even though microplots were inoculated with levels similar to the previous year.

In the strawberry experiment using *P. penetrans*, control plants had a significant amount of nematode reproduction with a mean of 109 nematodes/g root. No nematodes were obtained from Vydate treated plants. The lowest rate of butyric acid applies (0.01M) resulted in a three-fold reduction in nematode populations to 37 nematodes/g root. All chemical treatments were statistically similar to each other and statistically different from the control at  $P<0.001$ .

In both field seasons, ornamental hostas were used as a root-knot nematode host. Unfortunately, of the three varieties tested (Blue Cadet, Krossa Regal and Honeybells), none were susceptible to local populations of the nematode. Microplots were planted to hostas in both seasons using every treatment but nematode reproductive was never observed in controls of treated microplots. This result suggests that additional research must be undertaken to identify which nematode species are involved in the pathogenicity of hostas or whether specific biotypes are involved.

The experiment undertaken during the 2003 field season was repeated during 2004. Unfortunately, *P. penetrans* reproduction was extremely low in the greenhouse during the winter of 2003 and sufficient numbers of nematodes were not produced. Consequently, only the *M. hapla* portion of the experiment could be repeated. Results were very similar to those from the 2003 field season, demonstrating identical trends, although nematode numbers across all treatments were lower.

Weed data was collected in June 2003, prior to planting and after chemical treatment to determine the effect of treatments on summer annual weeds. The 0.1M butyric acid treatment provided 95.2% weed control and was statistically superior to all other treatments. The 0.05 M butyric acid and propionic acid treatments provided 76.1 and 70.1% weed control and were

statistically identical. The 0.01 M butyric acid, vydate and control treatments provided 7, 5.3 and 3.1% weed control and were statistically identical. Similar results were obtained from weed data collected during 2004, but on half as many replicate, resulting in less statistical significance.

In the Fall of 2002 a greenhouse experiment was set up to determine how significant a role pH plays in the activity of organic acids against plant-parasitic nematodes. Greenhouse pots were inoculated with root-knot nematodes and treated with four levels of propionic acid (1M, 0.5M, 0.1M and 0.05M) and one level of HCl (2.16 ml 3.7% to mimic pH levels in the 0.5M propionic acid treatment). After twelve weeks plants were harvested and nematode levels were measured. Control plants had a mean of 14,173 eggs/g root, statistically higher than all other treatments. HCl treated pots had a mean of 5,434 eggs/g root and were statistically less than the control plots. The 1M treatment killed all plants but the 0.5M, 0.1M and 0.05M treatments all had statistically fewer nematodes than the other two treatments (0, 576 and 0 eggs/g root, respectively) and were statistically identical. The experiment was repeated in the Fall of 2005 and similar results were obtained. This experiment clearly demonstrates that pH is only partly responsible for the nematicidal activity of organic acids.

## Nematode Biological Control

The northern root-knot (RKN, *Meloidogyne hapla*) nematode is among the persistent problems in vegetable and ornamental production systems in temperate climates. Without resistant cultivars and pending loss of methyl bromide (MBR) and few sustainable alternatives available, the vegetable and nursery industries face many challenges in managing RKN and other soil pests and diseases. Because of the overlap of vegetable and ornamental production systems with field crops, Michigan growers are subjected to soybean cyst (*Heterodera glycines*) nematode-free certification. In addition its wide distribution, *H. glycines*'s high degree of parasitic variability presents multiple management challenges as well. In order to meet the challenges, a multi-dimensional approach to developing MBR alternatives is needed. This objective was initiated to do the ground-work of screening for the presence of RKN fungal parasites in Michigan and to test the reaction of RKN populations to *Hirsutella minnesotensis* (*Hm*, nematode parasitic fungus) found to parasitize soybean cyst nematode juveniles in over 14% of soils from the Midwestern USA.

The study focused on four main areas: **i)** field survey to determine the presence of *Hm* and RKN populations in selected Michigan nursery and vegetable production systems; **ii)** testing the response of regional greenhouse RKN populations from RI, CT, NY (Geneva [NYG] and Lyndenville [NYL]), MI and WI to *Hm* and N-Viro Soil® (NVS), a recycled municipal biosolid with nutrient and pH adjustment qualities and adverse effects on several important plant-parasitic nematodes; **iii)** testing the response of NVS on *H. glycines*; and **iv)** characterizing the pathogenecity and adaptation behaviors of the Michigan RKN field populations (isolates). All greenhouse experiments were conducted using sandy loam soil with pH 7.0, and tomato cv 'Rutgers' was the host in all, but the *H. glycines* experiments.

Following is progress by area of study. **i)** Two field surveys were conducted. In 2003, analysis of 48 soil samples collected from Michigan nursery (Hosta, Moonbeam, Artemisia, Ajuga) and vegetable (celery, corn, carrot and potato) production systems showed the presence of varying degrees of RKN, root lesion and other nematodes. While fungal parasitism of root-knot nematodes ranging from 1% to

95% was observed in 37% of the samples, only one sample had 39% parasitism by *Pasteuria* spp.. In 2004, 60 soil and root samples from Hosta and celery were analyzed and fungal parasitism was observed in the latter. The surveyed fields included sandy, sandy loam and muck soils (which represent most common soil types for Michigan agriculture) and have variable history and degree of pesticide use. No *Hm* was found. Several RKN populations isolated from the surveys are being used for further characterization of their parasitic and adaptive behaviors.

The effects of *Hm* SD3-2 against regional greenhouse RKN populations from RI, CT, NYG, NYL, MI and WI was investigated using 20-day-old tomato seedlings. Nematodes were inoculated at either 0 or 600 eggs of each nematode population separately mixed with either 0, 0.02, or 0.1 g fresh *Hm* mycelium /100 cm<sup>-3</sup> of soil in pots containing 500 cm<sup>3</sup> soil and maintained at 25±2 °C for two months. While all *M. hapla* populations were suppressed by *Hm*, the degree to which each population was affected varied slightly. Across fungal treatments and nematode populations, the fungus reduced total number of nematodes in roots by 61-98%, with the highest for NYG and RI, intermediate for NYL and CT, and lowest for MI and WI populations. The study demonstrated that *Hm* may be used as a potential suppressor of tested *M. hapla* populations.

The interaction effects of *Hm* and NVS on the regional greenhouse RKN populations from RI, CT, NYG, NYL, and MI was tested using tomato seedlings. Nematodes were inoculated with either 0 or 600 eggs of each nematode population separately mixed with either 0 or 0.1 g fresh *Hm* mycelium and 0 or 1 g of NVS /100 cm<sup>-3</sup> of soil in pots containing 500 cm<sup>3</sup> soil and maintained at 25±2 °C for 1 month. *Hm* at 0.1 g reduced nematode number by 31-83% across nematode populations in one test, but only slightly reduced densities of NYG and CT populations in another test. NVS reduced nematode number by 33-92% across populations in two repeated tests. The combination of the two agents resulted in greater nematode reduction compared with *Hm* alone, but not compared with NVS alone. Across all fungal and NVS treatments, reduction of nematode number was generally greater in NYG, CT, and RI than in MI and NYL populations. Thus, demonstrating that *Hm* and NVS may be used as a potential suppressor of tested *M. hapla* populations.

The effects of 0, 1, 2 and 4 g NVS /100 cm<sup>3</sup> of soil on the regional greenhouse *M. hapla* populations from RI, CT, NYG, NYL, MI was investigated. Using tomato seedlings, two greenhouse experiments (25 ± 2 °C), each with two blocks consisting of 96 (4 NVS x 6 nems x 4 reps) experimental units, were conducted for 30 (Exp. 1) and 90 (Exp. 2) days after inoculation with either 0 (control) or 3,000 eggs / 500 cm<sup>3</sup> of soil. There was consistent interaction between *M. hapla* populations and NVS treatment in both experiments, showing that NVS does not affect *M. hapla* populations in the same way. The NYG and NYL populations showed consistent decrease in population density with increasing NVS treatment in both experiments. Compared with the controls, there was no significant decrease or increase in the population densities of the CT, RI and MI populations with increasing NVS dose, indicating some level of suppression. The 2 g NVS/100 cm<sup>3</sup> of soil appears to be slightly better for plant growth than the other treatments. The study indicates that NVS application against *M. hapla* is likely to be dose dependent and site-specific.

Soil amendments are being considered either as stand-alone and/or as part of integrated management approaches to deal with *H. glycines*'s complex biology. In three greenhouse experiments, the effects of 0, 1 or 4 g NVS per 100 cm<sup>3</sup> of soil on three *H. glycines* populations (GN 1, GN 2 and GN 3) was investigated using Round-up Ready<sup>®</sup> soybean (DSR-221) seedlings over 557 ± 68 degree-days (base 10 C). In the new *H. glycines* classification system, GN 1, GN 2 and GN 3 correspond to HG type 2, 1.2, and 0, respectively. Among the *H. glycines* populations, GN 3 generally infected the most and GN 1 the



least, suggesting that the populations possibly differ in their adaptation to the soil environment. There was variable response at 1 g NVS whereas 4 g NVS/100 cm<sup>3</sup> soil treatment was more uniformly effective in percent suppression of the total numbers of *H. glycines* cysts and other life stages of all three populations in roots. However, the higher dose was more toxic to the plants than the lower dose. The *H. glycines* populations responded similarly to high NVS dose but not at low dose, suggesting that effective NVS treatment may be site-specific.

Based on differences in field characteristics in the survey (i), *M. hapla* field populations from three nursery (1, 2 and 3) and one (4) vegetable field were selected for pathogenicity and adaptive behavior characterization. Fields 1, 2, 3, and 4 had loamy sand, sandy, sandy, and muck soils with pHs of 7.15, 6.56, 7.43, and 6.30, respectively, and significantly different from one another. The nursery fields showed significant imbalance in P, Ca and Mg while the vegetable field was nutritionally richer than the nursery fields. Nematode community structure analysis was done in Fields 1 and 4 as a measure of biological and ecological differences between the extremes of the field conditions. More herbivores and fungivores and less bacteriovores were found in Field 1 than in Field 4, indicating ecological differences between the two soil types. The four *M. hapla* field isolates were inoculated into tomato cv 'Rutgers' at 0 or 600 eggs with either 0, 1 and 4 g NVS/100 cm<sup>3</sup> of soil in three greenhouse experiments (28 ± 2 °C) for one month. With or without NVS, the population from the vegetable field was the least pathogenic. Populations densities of four isolates was inversely related to the field soil pHs. The results suggest that *M. hapla* populations may not respond similarly to MBr alternatives, challenging the "one-option-fits-all management approach". Furthermore, the data suggest that there may be soil physio-chemical bases to the differences in pathogenicity among the field isolates. Hence, identifying the possible mechanisms of different responses will be helpful to avoiding wrong conclusions and may be costly management decisions as well.

The study establishes valuable data base on: **a)** presence or absence of fungal nematode biocontrol agents; **b)** what potential biological and abiotic soil amendment options the nursery and vegetable industries may test against RKN and *H. glycines* infestation; **c)** possible ecological basis to RKN pathogenicity differences; and **d)** avoiding the pitfalls of one-option-fits-all management approach through exploiting our ability to understand nematode parasitic variability.

## **Weed Management Alternatives**

In the 3.2A field experiment associated with the first methyl bromide project, all of the treatments except idomethane plus chloropicrin at 200 lb/A, non tarped metham treatment *Lactuca serriola* under the Basamid treatment provided good weed control for up to 20 months. Phytotoxicity was not observed for *Echinops bannaticus* 'Blue Globe', *Lavandula angustifolia* 'Hidcote Blue', *Hosta* 'Twilight PP14040', *Artemisia schmidtiana* 'Silver Mound', *Chrysanthemum x superbum* 'Snow Lady' and *Coreopsis verticillata* 'Moon Beam'. Minor injury occurred with *Euphorbia polychrome*. The negative impact of Telone C-35 at 35 g/A on *Euphorbia* was the only effect of any of the treatments on plant dry weight.

**Field and container-grown conifer seedlings response to herbicides.-** Douglas Fir (DF) - (*Pseudotsuga menziesii*), Black Hills Spruce (BHS) – (*Picea glauca densata*), White Spruce (WS) – (*Picea glauca*), Colorado Blue Spruce (CBS) – (*Picea pungens glauca*), Eastern White

Pine (EWP) – (*Pinus strobus*), Canaan fir (CA) – (*Abies balsamea* var. *phanerolepis*) and Fraser fir (FF) – (*Abies fraseri*) were planted in the field and containers in East Lansing, MI, in 2003 and 2004.

Terbacil (1.12 kg/ha), imazaquin (0.42 kg/ha), flumioxazin granular (0.28 kg/ha), isoxaben (1.12 kg/ha) plus trifluralin (0.84 kg/ha), mesotrione (0.28 kg/ha), trifloxysulfuron (0.01 kg/ha) were sprayed over the top of the seedlings 3 weeks after planting. Rimsulfuron (0.025 kg/ha), imazapic (0.07 kg/ha), and lactofen (0.28 kg/ha) were added in the container experiment.

Weed control and tree injury were graded visually at 2 and 6 weeks after treatment on a scale from 1 to 10, meaning 1 no weed control or no crop injury and 10 complete weed control or dead crop. Tree size index ((height + width)/2) was measured at the end of each year.

A randomized complete block design was used for the statistical analysis with 3 replications for the field and 4 replications for the container experiment.

Terbacil (1.12 kg/ha), imazaquin (0.42 kg/ha), flumioxazin (0.28 kg/ha), isoxaben (1.12 kg/ha) plus trifluralin (0.84 kg/ha), mesotrione (0.28 kg/ha), and trifloxysulfuron (0.01 kg/ha) were safe on Colorado blue spruce (*Picea pungens glauca*) and eastern white pine (*Pinus strobus*) at 6 weeks after treatment in both years and to white spruce (*Picea glauca*) in 2003. Terbacil injured Douglas fir (*Pseudotsuga menziesii*), and Black Hills spruce (*Picea glauca densata*), Fraser fir (*Abies fraseri*), and Canaan fir (*Abies balsamea* var. *phanerolepis*). Mesotrione injured Douglas fir in 2004 and flumioxazin injured Fraser fir in 2004. Growth was only significantly different from the control in Black Hills spruce and Fraser fir treated with terbacil.

Terbacil, imazaquin, flumioxazin and mesotrione gave the best weed control. However, flumioxazin did not control common lambsquarters and redroot pigweed in 2003, and mesotrione did not control common purslane in both years. Trifloxysulfuron and isoxaben plus trifluralin had variable weed control among weeds species. Trifloxysulfuron controlled redroot pigweed in both years and common lambsquarters in 2004, while isoxaben plus trifluralin controlled common lambsquarters in 2004, eastern black nightshade, and broadleaf plantain in both years.

The same treatments as in the field experiment plus the additional treatments rimsulfuron (0.025 kg/ha), imazapic (0.070 kg/ha), and lactofen (0.28 kg/ha) were applied in the container experiment. Terbacil caused injury to all conifer species evaluated, except for Douglas fir and white spruce in 2003. Contrary to the field experiment, almost all treatments caused injury on Black Hills spruce and Colorado blue spruce. The high injury is explained by a higher herbicide concentration in the soil media contained in the pots, making the herbicides more available for being absorbed by the seedlings. Mesotrione injured white spruce in 2004 and lactofen injured eastern white pine in both years. The other herbicides did not cause significant injury to any species. Seedling growth was significantly reduced from the control in Canaan and Fraser fir treated with terbacil. All treatments reduced the number of annual sowthistle, fall panicum, and carpetweed.

Herbicides are effective in controlling annual weeds in conifer seedlings in containers and the field. Use of herbicides should save conifer seedling growers over \$1000 per acre per year.

**Perennials crop response to herbicides.**-Ten ornamental species were planted in a clay loam soil in East Lansing, MI, on July 1, 2003. Each replication had 10 rows and each row was planted with one species. Distance between plants was 30 cm with 120 cm between rows and the row length was 40 m. Herbicide plots were arranged perpendicular to the plant rows and they were 1.5 m wide and 11 m long and each plot crossed the ten ornamental species. For the crop evaluation (injury and size index) a strip plot design with 4 replications was used and for weed control evaluation a randomized complete block design. Seven herbicide treatments (including control) were sprayed over the top of the plants on July 14, 2003. Flumioxazin granular was applied manually with a shaker.

Ornamental injury and weed control were evaluated visually 2 and 6 weeks after treatment on a scale of 1 to 10; with 1 indicating no injury or no weed control and 10 indicating a dead crop plant or 100% weed control. Plant size index was measured on November of each year using the following formula: (height + width)/2. In 2004, plants at the opposite end of the row planted in 2003 were used for the experiment.

Terbacil (1.12 kg/ha), imazapic (0.07 kg/ha), imazaquin (0.42 kg/ha), halosulfuron (0.035 kg/ha), flumioxazin granular (0.28 kg/ha), and the tank mix isoxaben (1.12 kg/ha) plus trifluralin (0.84 kg/ha) did not cause injury on Holly (*Ilex* 'Blue Prince'), white spruce (*Picea glauca*), Anlojap yew (*Taxus media*), and white cedar (*Thuja occidentalis*) at 6 weeks after treatment. However, terbacil injured Japanese barberry (*Berberis thunbergii* 'Burgundy Carousel'), Redosier dogwood (*Cornus stolonifera* 'Alleman's Compact'), winged euonymus (*Euonymus alatus* 'Chicago Fire'), panicle hydrangea (*Hydrangea paniculata* 'Kyushu'), Japanese spirea (*Spiraea japonica* 'Fire Light'), and Preston lilac (*Syringa x prestoniae* 'Donald Wyman'). Imazaquin, imazapic, and halosulfuron had variable injury results among years and species. Flumioxazin and isoxaben plus trifluralin were the safest treatments on all species, except isoxaben plus trifluralin slightly injured winged euonymus. Imazaquin and imazapic caused reduced plant size index in Redosier dogwood. Terbacil had the best broadleaf weed control at 2 and 6 weeks after treatment. Imazaquin and flumioxazin effectiveness was comparable to terbacil at 6 weeks after treatment, with some exceptions. Imazaquin did not control common lambsquarters in 2003 and flumioxazin was less effective in controlling common lambsquarters in 2003, and common groundsel and common chickweed in 2004. Imazapic gave poor redroot pigweed and common lambsquarters control in 2003, but gave good control in 2004. Imazapic controlled all weeds evaluated and had acceptable control of common purslane and common groundsel at 6 weeks after treatment. Halosulfuron provided variable weed control among weed species. It had the best control on common groundsel, curly dock, and redroot pigweed in 2004; however, it gave only fair control of other weeds. Isoxaben plus trifluralin gave good control of common lambsquarters and redroot pigweed in 2004; control of the rest of the weed species was variable. These herbicides have the potential to replace MB for preemergence weed control in these ornamental crops.

Herbicides are effective in controlling annual weeds in woody ornamental crops. Using herbicides in place of hand weeding should save Michigan nurseries over \$3,000 per acre per year.

## 2005 Methyl Bromide Project II. Status Report (G. W. Bird Nematology Laboratory)

Soil and root tissue samples for nematode community structure analysis were taken at the Southwest Michigan Horticultural Research and Education Center methyl bromide alternatives research site prior to application of soil pesticides, at-planting and at the end of the 2005 growing season. The assessment indicates that the soil at the site has poor biological structural and enrichment attributes and that three genera of phytopathogenic nematodes are present at high enough population densities to inhibit normal plant growth of some of the herbaceous and woody perennial ornamental cultivars. High concentrations of heavy metals are present. It is possible that this may have been at least partially responsible for the poor establishment of one or more of the test cultivars.

This is the first demonstration of soil biological structure and enrichment in relation to the production of herbaceous and woody perennial ornamentals in Michigan. The test is also an excellent example of high population densities of phytopathogenic nematodes associated with poor quality soil. This should serve as a catalyst to start the industry thinking about soil quality, which may be essential for development of sound alternatives to methyl bromide. The occurrence of high concentrations of heavy metals may also have an impact on the approach of the industry to soil management.

nd Creeping Phlox and treated with Vydate 2L at 2 lbs ai/acre in 25 gal wate

**Table 5. Properties of the soil fumigants (Prepared by Robert Uhlig, Graduate Research Assistant, Department of Horticulture, Michigan State University).**

Comon name	Trade name	Formulation	Status	Pest contro
Methyl Bromide	Brom-o-Gas	MB 98% + CP 2%	Registered	Nematodes
Methyl Bromide	Terro-gas-67	MB 67% + CP 33%	Registered	Nematodes
Metham sodium	Vapam	Sodium methyldithiocarbamate 42% + 58% inerts	Registered	Nematodes
Chloropicrin	Chloropicrin	Chloropicrin 99.5%	Registered	Nematodes
1,3-D	Telone II	1,3-Dichloropropene 97.5%	Registered	Nematodes
1,3-D	Telone C-35	1,3-Dichloropropene 65% + 35% CP	Registered	Nematodes
1,3-D	Telone C-17	1,3-Dichloropropene 83% + 17% CP	Registered	Nematodes
1,3-D	Telone EC	1.3-Dichloropropene 93.6 %	Registered	Nematodes
Dazomet	Basamid	Dazomet granular 99%	Registered	Nematodes
Methyl iodide	Methyl iodide	MeI 98% + CP 2%	Not registered	Nematodes
Methyl iodide	Methyl iodide	MeI 50% + CP 50%	Not registered	Nematodes
Ozone	-		Not registered	
Propylene oxide	-		Not registered	
Propargyl bromide	-		Not registered	
Sodium azide	Sep-100		Not registered	

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